Occurrence of the Spirochaete Genus Cristispira in Western Canadian Marine Bivalves

BY

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INTRODUCTION

THE PRESENCE OF A LARGE cristaferous unicellular organism in the digestive tract of molluscs has been known since Certes (1882) reported it in Ostrea edulis LINNAEUS, 1758. CERTES (1891) considered it a trypanosome. LAVERAN & MESNIL (1901) suggested it was a bacterium, while Perrin (1906) placed it among the Protozoa on the basis of the complex cytological organization. Swellengrebel (1907) first recognized the spirochaete affinities and was followed by Gross (1910) who erected the genus Cristispira to contain those forms characterized by an encircling crista and the relatively large size. Noguchi (1921) reported upon the distribution of the genus in molluscs collected at Woods Hole, Massachusetts, and Dimitroff (1926) reviewed the literature to that date. Berkeley (1959) first recorded the genus from Western Canada in the large clam Saxidomus giganteus (DESHAYES, 1841).

METHOD

As many individuals as possible of 63 species of bivalves collected throughout the year from the intertidal to 1700 meters were examined for the presence of *Cristispira*. Examination was made as quickly as possible; in no case more than 10 minutes after collection. One valve was removed and a hypodermic needle inserted dorsally into the stomach and a small amount of stomach contents aspirated. The digestive system was then dissected and duplicate smears taken from the stomach, crystalline style (if present), the style pouch or mid-gut, and the hind-gut. One set of smears was examined at once by means of transmitted and oblique illumination. The second set was fixed in absolute alcohol and stained in Giemsa solution.

The distribution of *Cristispira* within the substance of the style of *Saxidomus giganteus* was determined by sectioning frozen styles.

RESULTS

Twelve of the 62 species examined contained Cristispira. All of these were suspension feeders, generally belonging to stomach type V (Purchon, 1960). All those with Cristispira were intertidal dwellers excepting Diplodonta orbella which was collected in 10 m. Some species including Entodesma saxicola were found to host Cristispira in intertidal situations, but solitary representatives collected by means of SCUBA in 10 - 20 m were invariably free of the spirochaete. A similar situation exists in Compsomyax subdiaphana; examination of 79 specimens from various localities in the northern portion of the Strait of Georgia in 60 - 200 m failed to yield Cristispira, but in shallower water more southern representatives of the species were hosts (pers. comm., Dr. R. G. B. Reid, University of Victoria). Commercial and natural beds of the Pacific oyster (Crassostrea gigas) are frequent hosts; however, no individuals collected from the head of Pendrell Sound contained Cristispira.

Considerable variation in infection sites may be demonstrated. In Saxidomus the area of maximum infestation is in the style pouch and consequently adhering to the crystalline style. In winter the stomach is often completely free but in summer small isolated numbers of Cristispira may be present. In Tresus the stomach is the chief site and numbers may be found in the intestine and large numbers in the pallial cavity. Earlier papers suggest that the spirochaete is present within the style substance. Careful sectioning of frozen styles showed that in no case had Cristispira penetrated the style but merely formed a

coating on the surface. Adhering *Cristispira* can also be removed by washing the complete style in saline solution and blotting dry.

Numbers of *Cristispira* present vary seasonally, being much more abundant in summer when concentration in the order of 8×10^6 per ml stomach contents may be reached in *Tresus*.

Cristispira are variable in outline and until a culture technique is perfected, it may be prudent to avoid amendments or additions to the nomenclature. Classifications based solely upon external morphology are open to doubt as various histological procedures such as fixation and staining radically alter the appearance. The 7th edition of Bergey (1957) lists 3 species which adequately cover the morphological varieties present in British Columbia marine clams. The results of examination are listed in Table 1 and representatives of each occurrence have been assigned to one of 3 groups; this does not imply any systematic affinity but merely a general group with characters in common.

Туре	Similar to	Size	Characteristic
	Cristispira		
δ	C. balbianii (CERTES, 1882)	$40 - 140\mu$	Obtuse ends
β	C. anodontae (Kysselitz, 1906)	44 - 88μ	Pointed ends
α	C. pinnae (Gonder, 1908)	10 - 60μ	Blunt ends

DISCUSSION

Berkeley (1959) reported that the crystalline style of Saxidomus giganteus contained a large population of Cristispira except at the distal end which impinges against the gastric shield where food is broken down and subjected to enzymatic action of the disintegrating style. Previously Berkeley (1933) had demonstrated that one of the products of the oxidizing action of the crystalline style upon plankton was probably glucosone. Berkeley (1962) carried out experiments which showed that plankton extracts were toxic to Cristispira suspensions in 60 to 90 minutes and further investigated the toxicity of glucosone. The results showed that addition of 0.5% glucosone to an active suspension of Cristispira in sea water held at 5° C rendered inactive all the spirochaetes in approximately one hour. Glucosone has been shown to be toxic to many animals (BECKER & DAY, 1953). DEAN (1958) reported that extracts of style caused rapid dissolution of Cryptomonas, though some algae including Isochrysis remained unaffected after 72 hours. The lysolytic activity of the style is transitory and present only during active disintegration. LAVIN (1946) first reported cellulolytic activity in the styles of Mactra and Mya. NEWELL (1953)

supported the contention and demonstrated similar activity in Ostrea edulis and Mytilus edulis, suggesting that cellulosis might be associated with the presence of Cristispira. No Cristispira has been found in Bankia setacea, which would appear to cast doubts upon Newell's suggestion. There is no direct correlation of the occurrence of the spirochaete and species of bivalve, though it appears that closely grouped intertidal populations are the most frequently infected.

The interesting situation where Cristispira-free populations of Crassostrea gigas and Venerubis japonica occupy the head of Pendrell Sound might be explained by the pronounced halocline present in these waters. The summer months are characterized by surface waters of relatively low salinity (10% - 20%) which could have an inhibitory effect upon Cristispira. Clams and ovsters from the mid region of the Sound demonstrated an infection percentage of 20 - 50, while at the mouth and surrounding areas virtually 100% of susceptible bivalves are infected. A contributing factor might be found in the precipitous walls of the Sound, which offer few suitable habitats for interstitial bivalves which could act as a Cristispira reservoir. No difference in conditions between the infected and Cristispira-free bivalves could be found, an indication that the spirochaete is not an obligative part of the gut fauna. The presence or absence of Cristispira appears to have little effect upon the well-being of the host and should probably be regarded as a commensal organism.

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Species	Depth	Number examined	Number positive	Type	Species	Depth	Number examined	Number soitive	Type
Acila castrensis (Hinds, 1843)	146	25	0		Mya arenaria Linnaeus, 1758	Int.	33	0	
Astarte alaskensis DALL, 1903	180	7	0		Mytilimeria nuttallii Conrad, 1837	Int.	2	0	
Astarte esquimalti BAIRD, 1863	23	9	0		Mytilus californianus Conrab, 1837	Int.	14	0	
Bankia setacea (TRYON, 1963)	Int.	28	0		Mytilus edulis LINNAEUS, 1758	Int.	35	0	
Cardiomya californica (DALL, 1886)	275	14	0		Nemocardium centifilosum (CARPENTER, 1864)	95	8	0	
Cuspidaria pectinata (CARPENTER, 1864)	180	33	0		Nucula carlottensis DALL, 1897	280	4	0	
Cardita ventricosa Gould, 1850	165	9	0		Nuculana cellulita (DALL, 1896)	750	3	0	
Chlamys rubida (HINDS, 1845)	110	14	0		Ostrea lurida CARPENTER, 1864	Int.	25	21	δ, β, α
Chlamys hericius (Gould, 1850)	110	6	0		Pandora bilirata Conrad, 1855	365	==	0	
Clinocardium nuttallii (Conrad, 1837)	Int.	27	25	δ,α	Pandora filosa (CARPENTER, 1864)	410	7	0	
Compsomyax subdiaphana (CARPENTER, 1864)	60-200	79	0		Pandora grandis Dall, 1877	250	10	0	
Crassostrea gigas (THUNBERG, 1793)	Int.	25	22	δ, α	Panope generosa Gould, 1850	Int.	-	-	β, α
(Pendrell Sound)	Int.	10	0		Pecten caurinus Gould, 1850	105	5	0	
Cuspidaria apodema Dall, 1916	1700	7	0		Pododesmus cepio (GRAY, 1849)	Int.	33	0	
Cyclopecten carlottensis Bernard, 1968	1650	-	0		Poromya beringiana (DALL, 1916)	750	-	0	
Diplodonta orbella (Gould, 1851)	10	2	7		Poromya tenuiconcha Dall, 1913	1385	3	0	
Entodesma saxicola (BAIRD, 1863)	10	9	0		Propeamussium davidsoni (Dall, 1897)	825	9	0	
Entodesma saxicola (BAIRD, 1863)	Int.	4	4	8,8	Protothaca staminea (Conrad, 1837)	Int.	25	24	δ, β, α
Gari californica (Conrad, 1849)	18	7	0		Saxidomus giganteus (Deshayes, 1839)	Int.	37	37	8, 8, 0
Gari californica (Conrad, 1849)	Int.	14	0		Serripes groenlandicus (BRUGUIÈRE, 1789)	25	2	0	
Glycymeris subobsoleta (CARPENTER, 1864)	8	20	0		Siliqua patula (Dixon, 1789) 1	Int.	2	0	
Hiatella arctica (LINNAEUS, 1767)	40	9	0		Solemya agassizii Dall, 1908	320	8	0	
Hinnites multirugosus (GALE, 1928)	20	4	0		Solen sicarius Gould, 1850	Int.	-	0	
Kellia suborbicularis (Montagu, 1804)	40	7	0		Tellina carpenteri Dall, 1900	20	4	0	
Lucinoma annulata (Reeve, 1850)	114	18	0		Tellina salmonea (CARPENTER, 1864)	75	15	0	
Lyonsia pugetensis Dall, 1913	Int.	7	7	β, α	Thyasira bisecta (Conrad, 1849)	170	8	0	
Macoma brota Dall, 1916	35	4	0		Thyasira disjuncta (GABB, 1866)	200	27	0	
Macoma calcarea (GMELIN, 1791)	65	12	0		Tresus capax (Gould, 1850)	Int.	2	2	β, α
Macoma inflatula Dall, 1897	40	9	0		Tresus nuttallii (Conrad, 1837)	Int.	2	5	β, α
Macoma nasuta (Conrad, 1837)	Int.	3	0		Venerupis japonica (Deshayes, 1841)	Int.	25	22	8, 8, 0
Macoma secta (Conrad, 1837)	25	2	0		Yoldia ensifera Dall, 1897	450	22	0	
Modiolus modiolus (LINNAEUS, 1758)	Int.	4	0		Yoldia thraciaeformis (Storer, 1838)	100	5	0	
Modiolus capax (Conrad, 1837)	20	2	0		Zirfaea gabbi (Tryon, 1863)	Int.	47	0	

BERKELEY (1959) lists Siliqua patula as being host to Cristispira.
 This is an error of identification and the record should be referred to Solen.

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